REMARKS/ARGUMENTS

This amendment is filed in response to the Official Action mailed September 11, 2007 for the above-referenced application. Reconsideration of the application, as amended, in view of the remarks herein is respectfully requested.

The Examiner maintained the restriction requirement asserting once again that the only common technical feature is geldanamycin. Applicants submit that this is an inappropriate view of the commonality between the claims, and accordingly will be filing a petition for review of the restriction requirement.

Claim 7 has been amended in view of the objection, and claim 32 in view of the rejection under 35 USC § 112, second paragraph. In addition, the specification and claim 3 have being amended to correct typographical errors.

The Examiner also rejected claim 6 under 35 USC § 112, second paragraph, stating that the phrase "at least partially different" is indefinite. Applicants respectfully disagree. As explained on Page 6 of the application, hsp90 dependent activity refers to expression of proteins that is dependent on hsp90. Since there are numerous proteins that can be hsp90-dependent, a partial difference is a change in the expression level of some proteins, but not necessarily in all proteins. Applicants submit that this language would be understood by a person skilled in the art, having read the specification, and therefore that the claim is not indefinite.

The Examiner claims stand rejected under 35 USC § 103 as unpatentable over the combination of Gewirth et al, in view of Rosen et al, Chiosis et al, Devlin et al, Bennett et al. and Pagé et al. Applicants respectfully traverse this rejection.

On Page 5 of the office action, the Examiner characterizes the invention as having two parts: a first part reflected in claim 1 in which a competitive binding assay is performed and binding is determined by measuring a reduction in fluorescence, and a second part defined in claim 3 in which biological activity is tested. In making this characterization, the Examiner has left out elements of the claimed invention. Specifically, in the first assay, what is determinative of binding is **not** a reduction in fluorescence but a reduction in the **polarization** of the fluorescence.

Gewirth is silent as to polarization of the fluorescence, and instead measures intensity changes. There is no evidence in the record that 8-ANS, the compound used in Gewirth would exhibit changes in fluorescence polarization as required by the present claims when bound to hsp90. The Examiner apparently recognizes this difference (Page 6 of the office action), and on

Page 8 starts to talk about polarization of fluorescence. It is not clear whether these arguments are to be applied in the context of claim 1, or merely in the context of claims 14-16 and 32-38, which are referenced in the immediately preceding paragraph. In the interest of advancing prosecution, Applicants will respond here as if they applied to claim 1, since the rejection of that claim is admitted to be deficient without some additional references.

The Examiner states that fluorescence polarization assays have advantages, and have been shown by Bennett to work with protein analytes, and because of this she argues that substitution of one assay technique (fluorescence polarization) for another (simple fluorescence) would have been obvious. This argument is overly simplistic, and fails to take into account the actual teachings of the references.

At a first matter, Applicants submit that Bennett is largely irrelevant with to the present invention. Bennett recaps difficulties that exist when trying to use fluorescence polarization for large protein analytes and states that C reactive protein (CRP) appears to be an exception in this case. The difficulty arises when the analyte analog to which the fluorescent moiety is bound is very large (as in the case of protein) so that the relaxation time of the unbound fluorescent molecule-protein combination is long. This makes it hard to tell when it becomes longer as a consequence of further binding. This difficulty is not relevant to the present application, because the molecule to which the fluorescent moiety is bound is not the protein (hsp90) but a binding moiety which is relatively small, and in any event Bennett teaches CRP as a special case, and not a general method for overcoming this problem which might be extended to other proteins.

The cited Devlin reference gives a more complete summary of the mechanism of fluorescence polarization measurements. However, this summary focuses on the polarization aspects of the assay, and fails to give consideration to the complexity that is associated with fluorescence of bound molecules. The property of fluorescence in a molecule arises from the presence of excited states which return to a ground state by releasing energy in the form of a photon (the emitted fluorescence) instead of through vibrational dissipation. Molecules which are physically rigid such as conjugated ring structures are therefore more likely to be fluorescent than more flexible molecules which have more vibrational modes. Fluorescence intensity can be lost from a normally fluorescent label as a consequence of binding/coupling to other molecules that provide vibrational modes, or the spectrum of the emitted light can change for example as a consequence of the environment surrounding the molecules.

In Gewirth, the binding of the inherently fluorescent 8-ANS is said to be based on structural similarities to the natural adenosine ligand and the moiety is selected because it initiates a structural change in the HSP90. (¶ 273 and 441) Gewirth does not disclose a distinct

label associated with a separate binding moiety that imparts specificity to the target binding site. Thus, there is no labeled ligand per se, but rather a ligand that also happens to be fluorescent.

The fluorescent moiety in Gewirth undergoes a change in the spectral characteristics of the fluorescence as a consequence of the hydrophobic nature of the pocket into which it is bound. (Col 41, ¶ 452). Furthermore, the ability of the 8-ANS to bind is effected by the conformational state of the pocket and the amount of time (it binds better after heat shock or after a long time, ¶¶ 453-4). A fluorescent moiety which is attached to a ligand (and which will therefore not be inside the pocket) would not display the types of effects considered in Gewirth, but would be affected by the proximity of the protein and interacting amino acid side chains on the outside of the pocket. Nothing in the cited art allows prediction of how these materials would interact when the protein is hsp90 and the fluorophore is a molecule such as FITC or BODIPY.

Furthermore, the fact reported in Gewirth that changes in hsp90 conformation can occur in response to the binding of certain ligands adds a further potentially complicating factor which makes prediction of the usefulness of FP as an assay technique in evaluating binding moieties impractical. If the protein undergoes a conformation change when bound, it may become more difficult to exchange binding species in and out of that site. This would lengthen the time required for the assay, making it less useful.

In short, there are many factors to consider in assessing whether the present invention would have been obvious. These factors create room for experimentation, and not a true suggestion to make the present invention. This is not simply a case of using one type of fluorophore in place of another, because in Gewirth, the fluorophore is the ligand and the change that is monitored depends on the ability of the fluorophore to fit within the pocket of the hsp90 molecule. Thus, Applicants submit that the rejection of claim 1, and of the claim dependent thereon is in error and should be withdrawn.

Applicants further note that the Examiner has not directly addressed the limitation of claim 2 that the hsp90 is provided in the form of a cell lysate. The Examiner does refer to the mention of cell based screening assays in Gewirth, but these assays do not involve the use of 8-ANS or any fluorescence measurements. Indeed, ¶¶ 473-8 discuss how the protein was purified for use in the assay.

Moreover, 8-ANS would be considered unsuitable for an assay in a cell lysate because it is well known that 8-ANS and other dyes of this structure indiscriminately bind to hydrophobic patches on proteins. Thus, in a cell lysate environment, binding would not be limited to hsp90 and any observations would be meaningless.

As explained in the present application, (Page 21, Example 9) the ability to use cell lysates as the source of hsp90 is beneficial because of the reduction in costs. However, cell lysates include many natural binders of hsp90 (including ATP) as well as presenting the hsp90 as part of a natural multi-protein complexes. This provides advantages because it works, but there is noting in the art which suggests that it would. Therefore, Applicants submit that claim 2 is separately non-obvious over the cited art.

With respect to claim 9, which specifies that the fluorescently-labeled molecule is a labeled geldanamycin labeled at the C17 position, the Examiner has cited Rosen et al. but has argued only why this suggests certain cell types, and not how Rosen connects with the assay of Gewirth to suggest the limitations of claim 9. Clarification is requested.

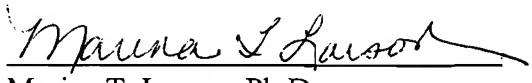
With regard to claim 17, the Examiner relies on unsupported allegations of knowledge in the art. Applicants submit that this is improper, and require that the examine rprovide the underlying basis (in the form of a reference or a declaration based on the Examiner's personal experience and expertise) so that Applicants can respond to facts in context rather than generalized allegations and conclusions. *In re Ahlert*, 165 USPQ 418, 420-21 (CCPA 1970).

With respect to claims 14-16 and 32-38, the Examiner states that several references disclose that "the geldanamycin 17-AAG is a ligand for HSP90." For clarity, Applicants point out that geldanamycin is a specific compound, not a class of compounds (geldanamycin is a type of ansamycin antibiotic). 17-AAG is a compound derived from geldanamycin.

Applicants have now added claims 41 and 42 which recite an additional component in the first assay, namely dithiothreitol (DTT). This amendment is supported in Examples 2, 8 and 16 as filed. Applicants have found that the presence of the DTT reducing agent results in an *in vitro* assay that better mimics the circumstances in a living cell such that there is better congruence between the *in vivo* bioaffinity and that measured in the assay. Nothing in the cited art discloses or suggests inclusion of DTT in a fluorescence polarization assay as claimed.

For the foregoing reasons, Applicants submit that the claims of this application should be allowed.

Respectfully submitted,



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